ISSN: 0975-833X

Available online at http://www.journalcra.com

International Journal of Current Research
Vol. 9, Issue, 05, pp.50184-50186, May, 2017

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

RESEARCH ARTICLE

ANTIMICROBIAL EFFICACY OF LYCOPENE COMPOUND AGANIST SOME PATHOGENS

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ARTICLE INFO

Article History:
Received 27th February, 2017
Received in revised form
05th March, 2017
Accepted 26th April, 2017
Published online 23rd May, 2017

Key words:
Tomato, Lycopene,
Pathogen.

ABSTRACT

Lycopene is a pigment principally responsible for the characteristic deep-red color of ripe tomato fruits and products. As a natural source of antioxidants, has attracted attentions due to its biological and physicochemical properties. In this study, tomato paste prepared from tomato cultivated in Dezfoul (Khozestan) was dehydrated with methanol, then lycopene was extracted with methanol-carbon tetrachloride mixture. Pure lycopene was obtained by twice crystallization of crude product from benzene through addition of boiling methanol. Further purification was achieved using column chromatography with alumina as the adsorbent. Effect of antimicrobial activity of lycopene compounds against some pathogens such as *S.aureus, E.coli, K.pneumoniae, P.aeruginosa, B.substilis* and fungi were analyzed including *Malassezia furfur* dermits.

Citation: Kavitha, G., Kanimozhi, K. and Panneerselvam, A.2017. "Antimicrobial efficacy of lycopene compound aganist some pathogens", *International Journal of Current Research*, 9, (05), 50184-50186.

INTRODUCTION

Recent epidemiological studies have suggested that the consumption of tomatoes and tomato-based food products reduce the risk of cancer (oral cavity, pharynx, esophagus, stomach, rectum, colon, urinary bladder, prostate and breast) in humans (Ferreira *et al.,* 2000; Tang *et al*., 2008 and Vaishampayan *et al*., 2007). This protective effect has been attributed to carotenoids, which are one of the major classes of phytochemicals in this fruit (Khachik *et al*., 2002). Carotenoids are a family of compounds of over 600 fat-soluble plant pigments that provide much of the color in nature. They are important nutritious for the human body owing to their provitamin A and antioxidant activities (Krinsky and Johnson, 2005). The most abundant carotenoid in tomato is lycopene, followed by phytoene, phytofluene, carotene, ß- carotene, neurosporene, and lutein. Lycopene, a red carotenoid pigment in tomatoes and tomato-based products, is an acyclic form of beta-carotene without pro-vitamin A activity. It has attracted substantial interest during recent times for its beneficial in reducing oxidative stressing coronary heart diseases and other chronic diseases (Rao and Agrawal, 2000; Rissanen *et al.,* 2002; Agarwal *et al*., 2001 and Upritchard *et al*., 2000). The antimicrobial activity of lycopene against oral pathogens and comparing to activity of commercial antibiotics. Lycopene is considered as one of the phytochemical, synthesized by plants and microorganisms. Lycopene is carotenoid, a natural pigment made by plants, which helps to protect plants from stress, and it also transfers light energy during photosynthesis. Lycopene is found in a number of fruits and vegetables, including apricots, guava, and watermelon.

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MATERIALS AND METHODS

Isolation of lycopene from tomato

Fifty grams tomato paste was dehydrated by adding 65 ml methanol. This mixture was immediately shaken vigorously to prevent the formation of hard lumps. After 2hr the thick suspension was filtered, the dark red cake was shaken for another 15 min with 75ml mixture of equal volume of methanol and carbon tetrachloride and separated by filtration. The carbon tetrachloride phase was transferred to a separator funnel, added one volume of water and shaken well. After phase separation the carbon tetrachloride phase was evaporated and the residue was diluted with about 2ml of benzene. Using a dropper, 1 ml of boiling methanol was added in portion, then crystals of crude lycopene appeared immediately and the crystallization was completed by keeping the liquid at room temperature and ice bath respectively. The crystals were washed 10 times using benzene and boiling methanol.

Antimicrobial Activity

For antibacterial and antifungal activity by preparing nutrient agar and potato dextrose agar respectively. Then dispense the

media into each of the petridish and allowed it to solidify. Transferred 1ml of 24 hrs old bacterial and fungal culture onto solidified plate and spread it with the help of sterile glass rod. After seeding with bacterial and fungal culture respectively, made the well at the centre of the media with the help of cork borer. Then transferred different concentration of lycopene to each of the well. Incubated the plates at 37^{o}C for 24 hrs for antibacterial activity and at room temperature for 2 to 4 days for antifungal activity observed plates for zone of inhibition, compared with the control and measure their diameter.

RESULT AND DISCUSSION

Lycopene is a natural source of antioxidants. The methanol extract of tomato studied for antibacterial activity by cup plate method. The antibacterial activity against *Bacillus substilus* showed better results. The growth of *Bacillus substilus* was inhibited about 25mm. zone of inhibition (Dhanawade and Sakhare, 2014). Effectiveness factor of the tomato is lycopene that possesses antibacterial and antifungal properties (Dahan *et al*., 2008; Rao, 2002). In the present study, the effect of antimicrobial activity of lycopene compound from *Lycopersicon esculentum* was more potential to inhibit the bacteria and fungi. It was 8, 10, 13 and 14 mm with 25, 50, 75 and 100µl of concentration lycopene compound treated respectively with bacterium of *S.aureus*. Lycopene interfere with the cell wall biosynthesis of *Staphylococuus aureus*. The destruction of cell wall occurs which ultimately leads to the cell death. Hence when lycopene is applied to the bacterial lawn of *Staphylococcus aureus* showed clear zone.

Table 1. Effect of Antibacterial activity of Lycopene compound against some bacteria

S.no	Name of the bacteria	Zone of inhibition (mm)			
		25 (µl)	50 (µl)	75 (µl)	100 (µl)
1	*B.substilus*	2	5	7	9
2	*E.coli*	10	13	15	19
3	*K.pneumoniae*	5	7	8	10
4	*P.aeruginosa*	4	5	8	11
5	*S.aureus*	8	10	13	14

Table 2. Effect of Antifungal activity of Lycopene compound against some fungi

S.no	Name of the fungi	Zone of inhibition (mm)			
		25 (µl)	50 (µl)	75 (µl)	100 (µl)
1	*A.flavus*	3	4	6	7
2	*A.niger*	5	7	9	11
3	*A.terreus*	3	6	7	9
4	*Candida* sp.	6	8	11	11
5	*Fusarium solani*	2	5	7	10
6	*Malassezia furfur*	5	7	9	11
7	*Microsporum* sp.	3	4	6	8
8	*Penicillin citrinum*	5	7	9	11

The efficiency of lycopene compound against *E.coli* was 10, 13, 15 and 19 mm with 25, 50, 75 and 100µl of concentration was treated but 100µl lycopene was highly suppressed. Accordingly the lycopene compounds showed moderate antibacterial activity observed. It was 5, 7, 8 and 10 mm zone of inhibition with 25, 50, 75 and 100µl of concentration of compounds against *K.pneumoniae* respectively. The maximum concentration (100µl) was excellent zone of inhibition recorded. While, *P.aeroginosa* was inhibited by lycopene compounds with 4, 5, 8 and 11 mm zone of inhibition with above mentioned concentration of samples were determined. Whereas *B.substilus* also more suppressive activity by lycopene compounds of 25, 50, 75 and 100µl of concentration was 2, 5, 7 and 9 mm minimum zone of inhibition was observed respectively (Table 1). *Salmonella typhimurium, Shigella flexneri, Staphylococcus aureus* and ETEC (Entero toxigenic *E. coli*) are the common pathogens those cause food borne intestinal disease (Haque *et al*., 2017). Lycopene have considerable antifungal activity against *Candida albicans*. Lycopene exerted potent antifungal activity on *Candida albicans* by causing significant damage to the cell membranes of the yeast hence clear zone was observed (Vaishampayan *et al*., 2007). In this study the antifungal activity of lycopene compounds from *L.esculentum* against fungi including demits were analysed. According to the different concentration of lycopene compound such as 25, 50, 75 and 100µl were 5, 7, 9 and 11 mm zone of inhibition of *A.niger* recorded respectively. The *A.flavus* was 3, 4, 6 and 7 mm zone of inhibition with concentration of 25, 50, 75 and 100µl of lycopene compounds respectively.

In the case of *A.terreus* inhibition zone was 3, 6, 7 and 9 mm zone of inhibition with above mentioned lycopene compounds observed respectively whereas *Fusarium solani* growth also determined with the suppression of lycopene compound was 2, 5, 7 and 10 mm zone of inhibition by 25, 50, 75 and 100µl of concentration of lycopene compounds. *Penicillium citrinum* was 5, 7, 9 and 11 mm inhibited with respective concentration of compounds. The *Candida* sp, *Microsporum* sp. and *Malassezia furfur* was inhibition by lycopene compounds of 25, 50, 75 and 100µl concentrations tested. The higher concentration 100µl was highly suppressiveness from these experiments. It was 6, 8, 11 and 11 mm zone of inhibition with 25, 50, 75 and 100µl of concentration of lycopene compounds were analyzed. In the case of *Microsporum* sp. the zone of inhibition was 3, 4, 6 and 8 mm with above mentioned concentration produced control measures, whereas *Malassezia furfur* dermits also controlled by lycopene compounds of 25, 50, 75 and 100µl with 5, 7, 9 and 12 mm zone was observed respectively (Table 2). It may be concluded that the lycopene compounds showed antimicrobial effect against *E.coli* and *Candida albicans* it may be as an antimicrobial agents against pathogens.

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